# RESPONSE TO REVACCINATION WITH TYPE III ORAL POLIOVIRUS VACCINE ( SABIN )

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# Response to Revaccination with type III Oral Poliovirus Vaccine (Sabin)

by

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## Introduction

In 1954, the first large scale trial of formalinized poliovirus vaccine was a major breakthrough in the eventual control of paralytic poliomyelitis.(1) Indeed, with the widespread use of mass immunization of the population in this country, the yearly rate of paralytic poliomyelitis has clearly been reduced. Morbidity statistics have shown that the number of paralytic poliomyelitis cases in the United States, which numbered approximately 15,000 annually during the period 1951-1955, dropped sharply between 1956 and 1959 to approximately 5,100 cases per year.(2) However several epidemics in 1959 halted this steady decline of paralytic cases which had been observed between 1956 and 1958.

One of the most important reasons for the recent increase in clinical poliomyelitis is that the population at large has failed to utilize the available vaccines.

Formalinized vaccines offer protection against the paralytic complication of a poliomyelitis infection by inducing the production of a type specific neutralizing antibody. However, it has been shown (3,4,5) that individuals having high levels of neutralizing antibody induced by formalinized vaccine, are still susceptible to alimentary

infection with poliovirus,and such subjects exhibit prolonged fecal excretion of poliovirus.

An inapparent natural infection with poliomyelitis virus, on the other hand, induces permanent protection against the paralytic disease even though the neutralizing antibodies may be at a relatively low level(6). When naturally immune individuals, particularly adults,(4) become reinfected, the period of fecal excretion of poliovirus is relatively short.(7,8) It has been suggested, therefore, that a local tissue factor in the epithelium of the intestinal tract of naturally immune individuals may play a role in protecting against reinfection. Efforts to produce a vaccine which would duplicate the natural processes of immunity without the risk of subsequent paralysis have led to numerous studies with both naturally occuring and experimentally produced attenuated strains of polioviruses.

Koprowski et al. were the first to administer a live rodent adapted poliovirus to volunteers in 1950.(9) In 1954, Sabin et al (10) found that polioviruses, after a number of rapid passages in monkey kidney tissue cultures and purification by the terminal dilution method, could be rendered almost completely avirulent for monkeys. In 1955 Sabin tested these strains of attenuated poliovirus in chimpanzees and human volunteers (11,12) and found that they produced an inapparent alimentary infection, in both;i.e.,without evidence of any associated paralysis. In these experiments the polioviruses which were recovered from the feces were found still to

be avirulent when innoculated intracerebrally into monkeys.

Koprowski et al. (13,14) fed attenuated types I and II poliovirus vaccines simultaneously to large numbers of nonimmune children and separately to a group of young infants. None of these subjects contracted clinical poliomyelitis and all developed homotypic antibodies. Neither injections of gamma globulin nor the presence of passively transfered antibodies affected the course of the inapparent alimentary infection.

Larger field trials with attenuated poliovirus vaccines have subsequently been conducted in various parts of the world.

Over 200,000 inhabitants of the Belgian Congo and Ruanda-Urundi were vaccinated with an attenuated type I poliovirus by Courtois et al. (16) in 1958. None of these individuals developed clinical poliomyelitis after the virus feeding. Subsequent to vaccination of all community members during four different outbreaks of poliomyelitis, no new cases of paralysis were reported.

Early in 1959 Sabin's strains of poliovirus vaccines were used in mass vaccination programs in the U.S.S.R. (17) Approximately 15,000,000 people were vaccinated and followed for a period of 6-9 months. During at least this surveilance period the vaccines seemed safe both for the vaccinees and for their communities.

The orally vaccinated republics experienced, during 1959, a marked reduction in paralytic cases, suggesting that the vaccine played a significant role in reducing the incidence of clinical poliomyelitis. In Tashkent vaccination was carried out in the midst

of an epidemic. The short duration, the sharp drop in cases of paralysis, and the relative immunity of the vaccinated as to non-vaccinated children all suggest that the live vaccine modified the course of the epidemic. Subsequently, in the U.S.S.R., more than 70,000,000 people have received the oral poliomyelitis vaccine. The very low incidence of clinical poliomyelitis in 1960 is encouraging.

Further understanding of the immune mechanisms of poliomyelitis infections is of major concern at present.

Experimental work in chimpanzees by Bodian et al. (18,19,20) and Howe (15,22) suggests that the level of serum neutralizing antibodies is of major importance in limiting the spread of ingested virus, and in high titers may even limit alimentary multiplication. In his earlier work Bodian et al. found that antibody alone could prevent viremia and paralysis after large amounts of peripherally innoculated poliovirus. The presence of such antibody, however, did not apparently affect fecal virus excretion nor did it prevent active antibody response subsequent to oral feeding of poliovirus. Since he found (23) large amounts of poliovirus in the tonsils and Peyer's patches but not in adjacent ileum of infected chimpanzees, Bodian postulated (21) that poliovirus multiplication primarily occured in these lymphoid tissues and subsequently was blood-borne to other areas of the body including the central nervous system. This systemic spread of poliovirus, he believed, could be prevented by relatively low levels of circulating homotypic antibodies.

On the other hand, Paul, Horstmann, (24) Neiderman, (25)

Sabin, (26) Fox (5) and others believe that while adequate levels of serum antibodies prevent or modify the paralytic complication of a poliomyelitis infection, the primary site of viral multiplication is the wall of the alimentary tract. As postulated by these workers, virus multiplication takes place within the superficial mucosal cells and/or the superficial lymphatics, the infective virus spreading from cell to cell until previously infected sites are encountered. Such an infection produces a local immunity which subsequently acts as a first line of defense in preventing reinfection with a homotypic poliovirus. Massive amounts of virus, however, can overwhelm this type of local intestinal immunity.

The present investigation is a continuation of studies of local immunity within the intestinal tract of man, which were begun by this department in 1956.(27)

These studies were conducted in a group of mentally retarded children and young adults who live in close association in a "closed"* institutional environment.

In the first trial, which was carried out in November, 1956, 20 children, aged 4-24, were fed a large dose of the L Sc strain of type I poliovirus vaccine. Of these subjects, 10 had type I antibodies induced by killed virus vaccine (the V group), and 10 had antibodies from "X" number of previous type I polio infections (the N group).

It was observed that the V individuals were much more readily infected than similar groups of N individuals. This was evidenced

*The children rarely leave the cottage-like-dormitory in which they live.

by prolonged virus excretion and sharp increases in neutralizing antibody levels of the V individuals, whereas the N subjects demonstrated relatively brief fecal excretion of virus and generally insignificant antibody responses.

On March of 1957, a second trial was initiated in which 11 individuals were refed the same large dose of the L Sc type I poliovirus. It was observed that after one alimentary infection with an attenuated poliovirus the V group responded in a manner very similar to the N group; i.e., they had shorter periods of fecal virus excretion and less striking antibody responses.

The effect of dosage in reinfecting the alimentary tract was determined in a third trial in November 1957. Four groups of 4 individuals were refed graded doses (ranging from $10^{7.4}$ TCD50 to $10^{2.4}$ TCD50) of the same type I poliovirus used previously. With a high dose of virus most individuals, both N and V, excreted significant amounts of virus and experienced at least a three fold increase in homotypic antibody titer. Lower doses of virus produced only isolated episodes of fecal virus excretion with no associated antibody increase,- see TABLE 5.

In April of 1958 a fourth trial, (28) similar in most respects to the first trial with attenuated type I poliovirus, was carried out with Sabin's KP-34 strain of type III attenuated poliovirus vaccine. A dose of $10^{4.5}$ TCD50 was fed a group of 6N and 6V individuals, aged 6-11 years. Regardless of the original immune status or of the level of homotypic antibodies, all 12 individuals

| DOSAGE | DAYS NO. | 1 | 3 | 5 | 7 | 9 | 11 | 13 | 21 | 28 | 38 | 45 | 56 | AB RISE |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| $10^{7.4}$ | 504 | ● | ● | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| | 513 | ● | ● | ● | ● | ● | ● | 0 | 0 | 0 | 0 | 0 | 0 | + |
| $TCD_{50}$ | 517 | ● | ● | ● | ● | ● | ● | ● | ● | 0 | ● | 0 | 0 | + |
| | 533 | ● | ● | 0 | 0 | ● | ● | 0 | ● | 0 | 0 | 0 | 0 | + |
| $10^{5.4}$ | 501 | ● | ● | ● | 0 | 0 | 0 | ● | 0 | ● | ● | 0 | ● | + |
| | 540 | ● | ● | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| $TCD_{50}$ | 550 | ● | ● | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| | 553 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| $10^{3.4}$ | 514 | 0 | ● | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| | 526 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| $TCD_{50}$ | 558 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| | 570 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| $10^{2.4}$ | 503 | 0 | 0 | 0 | 0 | ● | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| | 527 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |
| $TCD_{50}$ | 557 | 0 | 0 | 0 | 0 | 0 | ● | 0 | 0 | 0 | 0 | 0 | 0 | - |
| | 571 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | - |

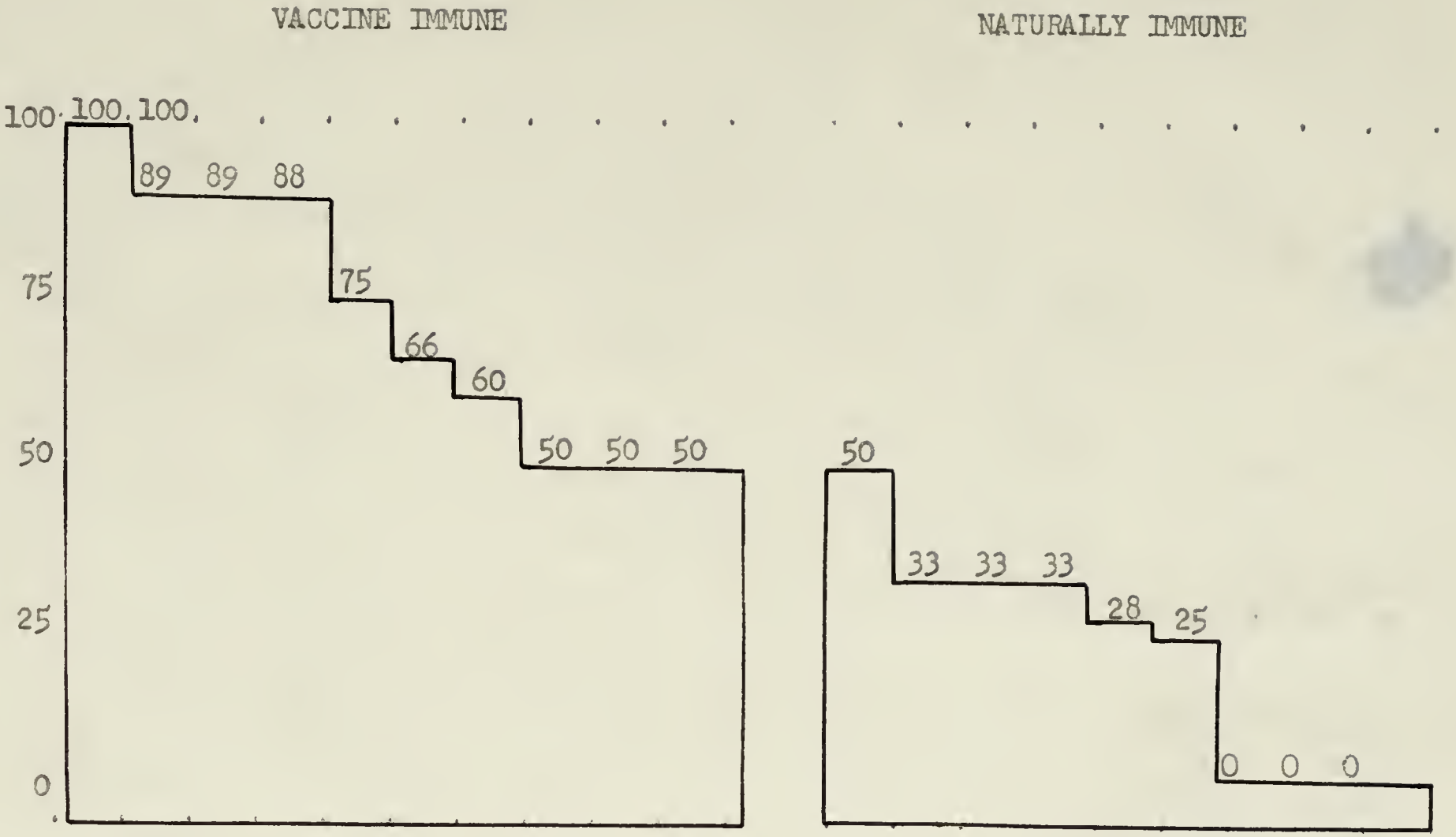
● POSITIVE STOOL
0 NEGATIVE STOOL

TABLE 5.- RESPONSE OF 16 PREVIOUSLY INFECTED CHILDERN TO FEEDING OF FOUR DIFFERENT GRADED DOSES OF L Sc TYPE I ATTENUATED POLIOVIRUS VACCINE.

| | | |

excreted significant amounts of fecal virus and/or experienced a marked antibody rise. Poliovirus excretion averaged 3 weeks in the N group and 5 weeks in the V group. Tables I and II compare these results with those of the first type I trial. All V individuals fed type I attenuated poliovirus responded with a significant increase in antibody titer while only 2 of the 9 in the N group experienced a similar antibody response. In contrast, substantial antibody increases were apparent in 5/6 individuals in both the N and the V groups following administration of type III poliovirus. The active immune response experienced by the naturally immune individuals fed the type III poliovirus suggested that immunity induced by an infection with type III poliovirus might in some way differ from that induced by an infection with the type I poliovirus. Furthermore, it was observed, when antibody responses are correlated with the number of positive stools (Tables I, II, and III), that the N subjects fed the type I poliovirus vaccine were the only group which was not heavily infected during the first 10 post-feeding days.

The present study is concerned with determining the immune responses in humans following a second feeding of Sabin's type III oral poliovirus vaccine. A titration type of trial in which graded doses of virus were fed was designed in an attempt to obtain quantitative data comparing, if possible, the ease of reinfectivity to the prefeeding homotypic antibody titer, and to measure the duration and amount of poliovirus excreted in the stools of both N and V individuals who have had one previous alimentary infection with the attenuated

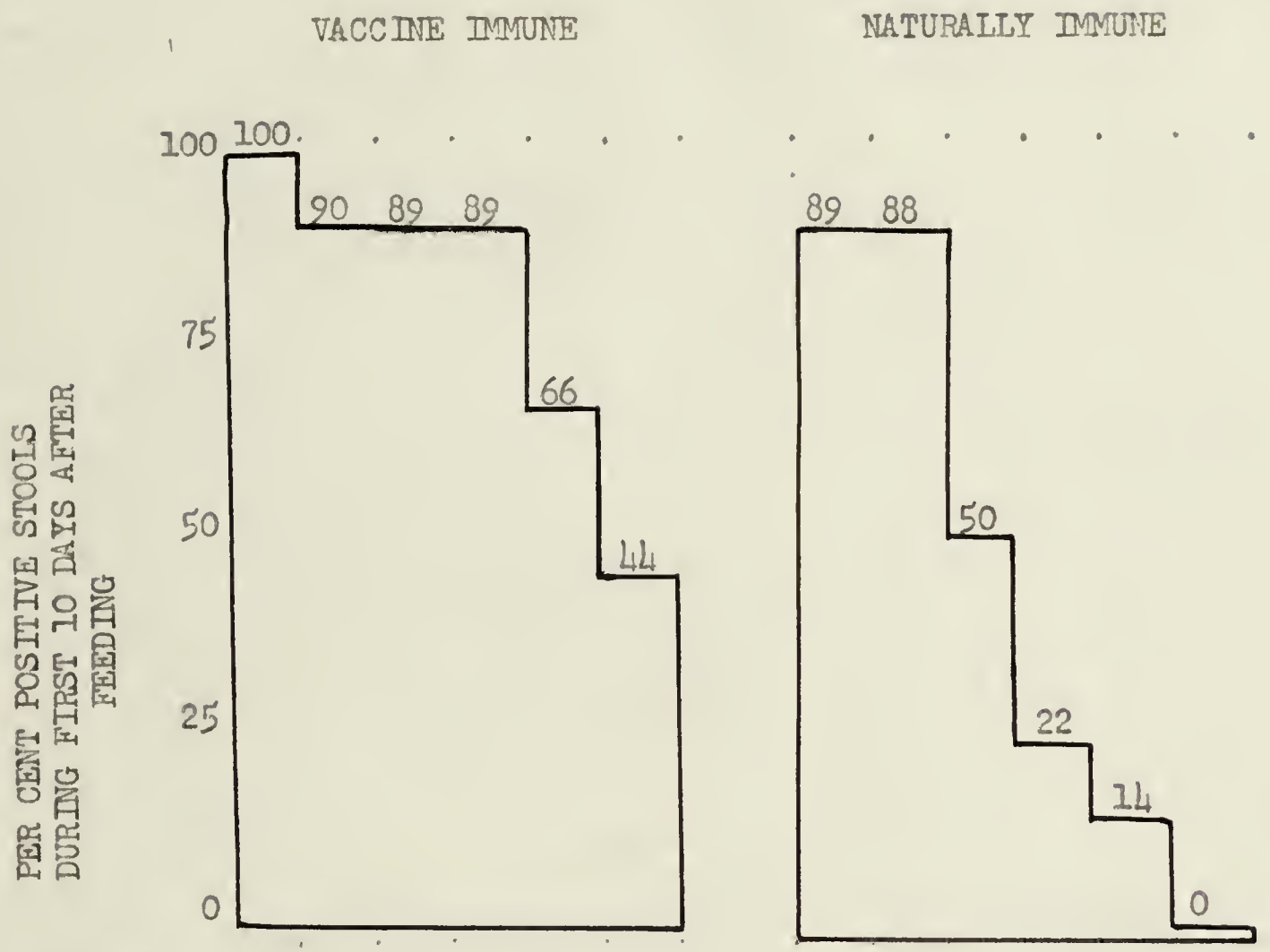



| | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| # POSITIVE STOOLS / # STOOLS COLLECTED | 7/7 | 4/4 | 8/9 | 7/8 | 3/4 | 2/3n6/10 | 3/6 | 3/6 | 4/8 | | 2/4 | 2/6 | 2/6 | 1/3 | 2/7 | 1/4 | 0/3 | 0/3 | 0/5 |
| SUBJECT # | 501 | 517 | 503 | 526 | 546 | 513 | 514 | 505 | 527 | 540 | 550 | 553 | 571 | 558 | 561 | 542 | 504 | 533 | 570 |

TABLE 1.- INDIVIDUALS FED $10^{7.4}TCD_{50}$ OF SABIN'S TYPE I ATTENUATED POLIOVIRUS VACCINE, NOVEMBER, 1956.

TABLE 2.

INDIVIDUALS FED $10^{4.5}$ $TCD_{50}$ OF SABINS TYPE III ATTENUATED POLIOVIRUS VACCINE, APRIL, 1958




| # POSITIVE STOOLS / # STOOLS COLLECTED | 9/9 | 9/10 | 8/9 | 8/9 | 6/9 | 5/9 | 8/9 | 7/8 | 5/10 | 2/9 | 1/7 | 0/8 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| SUBJECT # | 540 | 547 | 520 | 571 | 528 | 553 | 506 | 521 | 511 | 526 | 514 | 516 |

TABLE 3.

PREFEEDING ANTIBODY TITRE AND ANTIBODY INCREASE IN INDIVIDUALS FED EITHER SABIN'S TYPE I OR TYPE III ATTENUATED POLIOVIRUS VACCINE.

| | | SUBJECT # | PREFEEDING AB TITRE | MAXIMAL FOLD INCREASE IN AB TITRE |
|---|---|---|---|---|
| FED TYPE I | VACCINE IMMUNE | 503 | 64 | X5 |
| | | 513 | 64 | X6 |
| | | 526 | 64 | X3 |
| | | 546 | 64 | X4 |
| | | 501 | 32 | X5 |
| | | 505 | 16 | X4 |
| | | 540 | 16 | X6 |
| | | 514 | 0 | X10 |
| | | 517 | 0 | X8 |
| | | 526 | 0 | X8 |
| | NATURALLY IMMUNE | 558 | 1024 | X2 |
| | | 504 | 512 | X0 |
| | | 533 | 512 | X1 |
| | | 570 | 512 | X0 |
| | | 571 | 512 | X1 |
| | | 542 | 64 | X0 |
| | | 550 | 64 | X2 |
| | | 553 | 64 | X4 |
| | | 561 | 32 | X4 |
| FED TYPE III | VACCINE IMMUNE | 540 | 256 | X2 |
| | | 547 | 256 | X3 |
| | | 520 | 256 | X4 |
| | | 571 | 256 | X4 |
| | | 528 | 16 | X5 |
| | | 553 | 64 | X5 |
| | NATURALLY IMMUNE | 506 | 1024 | X1 |
| | | 521 | 64 | X3 |
| | | 511 | 256 | X4 |
| | | 526 | 64 | X6 |
| | | 514 | 64 | X4 |
| | | 516 | 64 | X3 |

type III poliovirus.

## METHODS AND MATERIALS

This trial was carried out at the Southbury Training School where the Yale Poliomyelitis Study Unit has previously conducted a series of trials with Sabin's type I and type III attenuated poliovirus vaccines. The risk of encountering wild enterovirus infections, including polioviruses, in this type of "closed" cottages where the present trial was conducted is very slight. In two "closed" cottages, 20 children ranging in age from 9-14 years, who had previously received Sabin's attenuated type III poliovirus vaccine, either by direct feeding or by contact infection in April, 1958, were fed graded doses of the same type III oral poliovirus vaccine. All of these children have received liberal amounts of formalinized vaccine and were last immunized on October 1, 1959 and January 10, 1961. Prior to the 1958 trial one half of these children's homotypic immunity had been induced by only killed poliovirus vaccines. At present their local intestinal immunity is due to one infection with the type III oral poliovirus vaccine (Sabin). The homotypic antibodies of the other one half of the present trial group have been induced both by natural type III polio infections and the killed poliovirus vaccine. Their local intestinal immunity is due to N numbers of natural type III poliovirus infections and one infection with the type III oral poliovirus vaccine (Sabin). These groups of individuals will be referred to as the V or N group respectively.

Virus:

Dr. Albert B. Sabin kindly made available the attenuated type III poliovirus, KP-34, used in the previous 1958 experiment.(28)

Using monkey kidney tissue culture tubes (10 tubes per log step) the virus titred, as calculated by Kaerber's method, $10^{5.2}$ TCD50 per 0.1 ml prior to feeding. The graded doses of virus were mixed with a sugared water to insure assimilation of the entire dose, and fed by teaspoon approximately one hour before the noon meal.

Specimens:

Blood

Blood specimens were collected in vaccuum syringes prior to the virus feeding and on the 7th., 28th., and 42th. days thereafter. The serum was separated and frozen on the day of collection.

Neutralization tests were carried out by the pH colorimetric method as modified by Melnick and Opton (29), using disposable plastic panels, and versenized second passage monkey kidney cells. Sera were tested at 1:4, 1:8,1:16, 1:32, 1:64, 1:128,1:256, 1:512, 1:1024 and 1:2048, 1:4096 and 1:8192 using 1 cup per dilution.

Complement fixation tests (designated hereafter as CF) were set up on plastic plates, using the method of Fulton and Dumbell (30) as modified by Black and Melnick (31). Tissue culture-grown antigens of type III poliovirus was used.

Stools

When possible daily stool samples were collected for the

first 10 days and biweekly thereafter for a period totaling six weeks. These were stored in the frozen state until prepared in the routine manner used in this laboratory (32). After thawing overnight, a 10 per cent stool suspension was made by mixing 3 gm. of stool with 27 ml. of sterile water in a pyrex bottle. After shaking for 1 hour on a mechanical shaker the suspension was transferred to a lusteroid tube and centrifuged at 3,500 R.P.M. for 1 hour. To the supernatant fluid 500 ug. of penicillin and 500 ug. of streptomycin per ml. were added; this constituted the inoculum and was stored at 4 degrees C. until innoculated shortly thereafter. Three oz. culture bottles containing trypsinized monolayer monkey kidney tissue grown in an out-growth medium containing 0.5 per cent lactalbumin hydrolysate, 2 per cent calf serum, 97.5 per cent Hank's salt solution and antibiotics were used through out.

After removing the media 2 ml of the 10 per cent stool suspension was innoculated into each tissue culture bottle. The bottles were incubated at 37 degrees C. for 1 1/2-2 hours, after which the stool suspension was removed and the cells washed twice with 4 ml. of Hank's balanced salt solution. Six ml. of a maintainance media (32) with twice concentrated bicarbonate was then applied. The bottles were read daily after the second day for 1 weeks and every other day thereafter for a period of 12 days.

All isolates were identified in neutralization tests set up against type III poliovirus as well as a pool of the 3 polio types (29). On those positive samples an aliquot of the supernate

of the original stool specimen was titrated in log dilutions (3 tubes per dilution) to determine the amount of virus excreted per gm. of stool.

Rectal Swabs

In those individuals from whom daily stool specimens were not obtained, rectal swabs were substituted. These swabs were tipped with cotton which was moistened with sterile 50 per cent glycerine before use. Each specimen was subsequently placed in a small vial containing 2 ml. of Hank's balanced salt solution. The vials were frozen at -20 degrees C. until prepared for innoculation as follows. The cotton was removed from each swab, then washed and squeezed dry 20 times in a 2 ml. syringe. The Hank's solution in which the specimens were stored was used for this procedure. This solution was then transferred to a lusteroid tube containing 1 ml. of Hank's with an added drop of 3.75 per cent $NaHCO_3$ and then centrifuged at 3,500 R.P.M. for 1 hour. To the supernatant fluid 500 ug. of penicillin and 500 ug. of streptomycin per ml. were added; this constituted the innoculum.

One ml. of this fluid was then innoculated in bottles as had been done with the stool preparations. The cells and fluid were then incubated for approximately 2 hours, after which time maintainance media (32) was added to the bottles without removing the innoculum or washing the cells. The bottles were first read on the 3rd. day and every other day thereafter for a period of 12 days. Positive samples were treated in the same way as the positive stool samples.

## RESULTS

In this trial, as in all the others, none of the children who received type III oral poliovirus vaccine (Sabin) developed any signs of clinical illness.

The preliminary results of stool excretion and complement fixing antibody titer (C.F.) are summarized in Table 4.

Three of 4 individuals who were fed the largest dose of virus excreted poliovirus on at least 2 occasions during the first postfeeding week. Four subjects fed the lower doses of virus excreted this virus less consistently and generally later, about the second postfeeding week, than those feed the largest dose of virus.

Five of the 7 subjects excreting virus had antibodies originally induced by Salk vaccination, with prefeeding titers ranging from 128-2048. The 2 originally naturally immune subjects who excreted virus had antibody titers of 2048 prior to this virus feeding.

The rise in C.F. antibodies in subjects No. 547 and No. 526 are suggestive of significant intestinal infection.

It was observed that 7 of 8 individuals with evidence of at least transient intestinal infection live in the same cottage.

| DOSAGE | SUBJECT NUMBER | ORIGINAL IMMUNE STATUS | PREFEEDING TYPE III AB TITRE | DAYS AFTER VIRUS FEEDING 3 | 4 | 5 | 6 | 7 | 14 | 15 | 16 | TITRE OF CF ANTIBODIES "PRE" | 1 WK | 4 WK |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| $10^{5.4}$ | 514 | V | 2048 | | + | | | + | | | - | 0 | 0 | 0 |
| | 511 | V | 128 | | + | | | + | | - | | 0 | 0 | 0 |
| $TCD_{50}$ | 545 | N | 2048 | | + | | | + | - | | | 0 | 0 | 0 |
| | 553 | N | 128 | | | - | | - | | - | | 0 | 0 | 0 |
| $10^{4.4}$ | 540 | V | 512 | | - | | | - | | | - | 0 | 0 | 0 |
| | 506 | V | 64 | - | | | | - | - | | | 0 | 0 | 0 |
| $TCD_{50}$ | 570 | N | 512 | | - | | | - | | | - | 0 | 0 | 0 |
| | 559 | N | 32 | | - | | | - | - | | | 0 | 0 | 0 |
| $10^{3.4}$ | 520 | V | 2048 | | | | | - | + | | | 0 | 0 | 0 |
| | 526 | V | 512 | | | | | + | - | | | 0 | 0 | 8 |
| $TCD_{50}$ | 503 | N | 2048 | | | - | | | | | + | 0 | 0 | 0 |
| | 557 | N | 128 | - | | | | - | - | | | 4 | 0 | 0 |
| $10^{2.4}$ | 547 | V | 2048 | | | | | - | - | | | 0 | 4 | 8 |
| | 516 | V | 8 | | - | | | - | | - | | 0 | 0 | 0 |
| $TCD_{50}$ | 501 | N | 1024 | | - | | | - | | - | | 0 | 0 | 0 |
| | 513 | N | 128 | | - | | | - | - | | | 0 | 0 | 0 |
| $10^{1.4}$ | 521 | V | 256 | | - | | | - | - | | | 0 | 0 | 0 |
| | 528 | V | 512 | | | | | - | + | | | 0 | 0 | 0 |
| $TCD_{50}$ | 527 | N | 8 | | - | | | - | - | | | 0 | 0 | 0 |
| | 568 | N | 32 | | | - | | - | - | | | 0 | 0 | 0 |

+ POSITIVE STOOL
- NEGATIVE STOOL
V VACCINE INDUCED ANTIBODIES
N NATURALLY ACQUIRED ANTIBODIES

TABLE 4.- REVACCINATION WITH TYPE III ORAL VACCINE (SABIN)
Responses of 20 children, aged 9-14, to the feeding of five different graded doses.

## DISCUSSION

The seemingly consistant pattern of excretion during the first postfeeding week in 3 of 4 individuals who were fed the largest virus dose in this trial suggest a reaction similar to that experienced by the subjects fed the largest dose of type I virus in the third type I trial (Table 5). Individuals fed the lower dose of type I poliovirus excreted this virus only sporadically (Table 5), and failed to show a homotypic neutralizing antibody response. The preliminary results of the present trial suggest a similar response in the individuals fed the lower doses of type III poliovirus (Table 4).

Similar responses to large feedings of both type I and type II poliovirus suggest that the relationship between immunity and the potential for reinfection in an individual, is relative, a certain level of immunity being overcome by a larger dose of virus.

At the highest virus dose both N and V individuals seemingly were intestinally infected, even with homotypic antibody levels as high as 2048.

In the 1958 type III trial, subject No. 504, aged 6 and naturally immune with a prefeeding antibody titer of 1024, excreted the greatest number (9/10) of positive stools during the first 10 post-feeding days. Even in the face of such extensive virus excretion a significant antibody increase could not be detected. All the other N subjects, none with titers higher than 256, experienced at least a 3 fold increase in antibody titer. This suggests that a ceiling antibody level is present that cannot be significantly raised even

by an intensive intestinal infection with the type III poliovirus.

If such is the case, one might anticipate that the individuals in the present trial with prefeeding antibody titer of 2048 (Subjects No. 514 and No. 548, Table IV) would not experience a significant homotypic antibody response even if found to excrete significant amounts of virus.

It is interesting to note that 5 of 8 subjects showing any evidence of infection (No.'s 514, 545, 520, 503 and 547) had prefeeding antibody titers of 2048. This suggests that with type III and unlike type II, circulating homotypic antibody plays little part in preventing virus multiplication within the intestinal tract. The sporadic pattern of excretion seen in all reinfected individuals except those fed the largest dose of virus suggests that with smaller amounts of virus a "local immunity" induced by previous intestinal infections plays a role in preventing virus multiplication in the intestinal tract.

Horstmann et al., in 1957, fed 4 naturally immune adults and 1 child with antibodies induced by killed vaccine, 2 different doses of the same type III oral poliovirus vaccine which was used in the present study. The results of this 1957 study are briefly summarized in Figure I.

| Virus Dose | Original Immune Status | Virus excreted in stools | Antibody rise |
|---|---|---|---|
| $10^{7.5}$ TCD50 | N | + | + |
| | N | + | + |
| $10^{4.5}$ TCD50 | N | + | + |
| | N | - | - |
| | V | + | + |

The reaction of the individuals fed the larger dose in her experiment seems similar to that of the N individuals in the type III Southbury trial, while the response of individuals to the lower dose of virus is more in keeping with the preliminary results of the present trial. This suggests that a massive enough dose may be able to overcome any degree of previous induced immunity and cause a significant intestinal infection.

## SUMMARY

Twenty children were fed graded doses of Sabin's oral type III poliovirus vaccine. None of the children receiving the vaccine showed any signs of clinical illness.

These results suggest that with type III as with type I poliovirus dosage is an important consideration in initiating re-infection.

High levels (2048) of homotypic antibody did not seen to play a role in preventing intestinal multiplication of the type III poliovirus.

These results indicate that a "local immunity phenomenon", present in individuals previously exposed to an alimentary infection with poliovirus, is effective in preventing reinfection with a homotypic poliovirus at lower virus doses, and that a massive dose of poliovirus may induce a significant intestinal infection reguardless of the previous immune status.

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